In the Claims

The claims are as follows:

1. (Original) An isolated nucleic acid molecule comprising a nucleic acid sequence encoding a fusion polypeptide comprising a reporter protein and at least two different heterologous protein destabilization sequences, which fusion polypeptide has a reduced half-life relative to a corresponding reporter protein which lacks the heterologous protein destabilization sequences or has a reduced half-life relative to a corresponding reporter protein which has one of the heterologous protein destabilization sequences.

2. (Original) An isolated nucleic acid molecule comprising a nucleic acid sequence comprising an open reading frame for a reporter protein and at least two heterologous destabilization sequences, wherein one of the heterologous destabilization sequences is a mRNA destabilization sequence and another is a heterologous protein destabilization sequence.

3. (Original) An isolated nucleic acid molecule comprising a nucleic acid sequence comprising an open reading frame for a luciferase and at least one heterologous destabilization sequence, wherein a majority of codons in the open reading frame for the luciferase are codons which are preferentially employed in a selected host cell.

4. (Original) The isolated nucleic acid molecule of claim 1, 2 or 3 further comprising a promoter operably linked to the nucleic acid sequence.

5. (Original) The isolated nucleic acid molecule of claim 4 wherein the promoter is a regulatable promoter.

6. (Original) The isolated nucleic acid molecule of claim 5 wherein the promoter is an inducible promoter.

7. (Original) The isolated nucleic acid molecule of claim 5 wherein the promoter is a repressible promoter.

8. (Original) The isolated nucleic acid molecule of claim 1 further comprising a heterologous mRNA destabilization sequence.

9. (Original) The isolated nucleic acid molecule of claim 2 or 8 wherein the mRNA destabilization is 3′ to the nucleic acid sequence.

10. (Original) The isolated nucleic acid molecule of claim 1 or 2 wherein the nucleic acid sequence encoding at least the reporter protein is optimized for expression in a host cell.

11. (Original) The isolated nucleic acid molecule of claim 1 or 2 wherein the reporter protein encodes a luciferase.

12. (Original) The isolated nucleic acid molecule of claim 1 wherein the reporter protein encodes a beetle luciferase.

13. (Original) The isolated nucleic acid molecule of claim 12 wherein the reporter protein encodes a click beetle luciferase.

14. (Original) The isolated nucleic acid molecule of claim 1 wherein the reporter protein encodes an anthozoan luciferase protein.

15. (Original) The isolated nucleic acid molecule of claim 3 wherein the heterologous destabilization sequence is a protein destabilization sequence.

16. (Original) The isolated nucleic acid molecule of claim 3 wherein the heterologous destabilization sequence is a mRNA destabilization sequence.

17. (Original) The isolated nucleic acid molecule of claim 1, 2 or 3 wherein nucleic acid sequence comprises SEQ ID NO:47, SEQ ID NO:48, SEQ ID NO:49, SEQ ID NO:66, SEQ ID NO:69, SEQ ID NO:70, SEQ ID NO:71, SEQ ID NO:72, SEQ ID NO:73, SEQ ID NO:74, SEQ ID NO:75, SEQ ID NO:76, SEQ ID NO:77, SEQ ID NO:78, SEQ ID NO:79, SEQ ID NO:80, or a fragment thereof that encodes a fusion polypeptide with substantially the same activity as the corresponding full-length fusion polypeptide encoded by SEQ ID NO:47, SEQ ID NO:48, SEQ ID NO:66, SEQ ID NO:69, SEQ ID NO:70, SEQ ID NO:71, SEQ ID NO:72, SEQ ID NO:73, SEQ ID NO:74, SEQ ID NO:75, SEQ ID NO:76, SEQ ID NO:77, SEQ ID NO:78, SEQ ID NO:79 or SEQ ID NO:80.

18. (Original) The isolated nucleic acid molecule of claim 1 further comprising a mRNA destabilization sequence.

19. (Original) The isolated molecule of claim 18 wherein one protein destabilization sequence is a PEST sequence.

20. (Original) The isolated nucleic acid molecule of claim 1 or 2 wherein one heterologous protein destabilization sequence is a PEST sequence.

21. (Original) The isolated nucleic acid molecule of claim 1 or 2 wherein one heterologous protein destabilization sequence is from the C-terminus of a mammalian ornithine decarboxylase.

22. (Original) The isolated nucleic acid molecule of claim 1 or 2 wherein one heterologous protein destabilization sequence is a mutant ornithine decarboxylase sequence.

23. (Original) The isolated nucleic acid molecule of claim 21 wherein the mutant ornithine decarboxylase sequence has an amino acid substitution at a position corresponding to position 426, 427, 428, 430, 431, 433, 434, 439 or 448 of murine ornithine decarboxylase.

24. (Original) The isolated nucleic acid molecule of claim 1 or 2 wherein one heterologous protein destabilization sequence is CL1, CL2, CL6, CL9, CL10, CL11, CL12, CL15, CL16, CL17 or SL17.

25. (Original) The isolated nucleic acid molecule of claim 1 or 2 wherein one heterologous protein destabilization sequence is at the C-terminus of the reporter protein.

26. (Original) The isolated nucleic acid molecule of claim 1 or 2 wherein one heterologous protein destabilization sequence at the N-terminus of the reporter protein.

27. (Original) The isolated nucleic acid molecule of claim 1 or 2 further comprising an ubiquitin polypeptide at the N-terminus of the fusion polypeptide.

28. (Original) The isolated nucleic acid molecule of claim 27 wherein one of the heterologous protein destabilization sequences is at the C-terminus of ubiquitin.

29. (Original) The isolated nucleic acid molecule of claim 28 wherein one of the heterologous protein destabilization sequences comprises a glutamic acid or arginine residue.

30. (Original) The isolated nucleic acid molecule of claim 10 which encodes a fusion polypeptide with a half-life of expression of about 20 minutes.

31. (Original) The isolated nucleic acid molecule of claim 10 which encodes a fusion polypeptide with a half-life of expression of about 30 minutes.

32. (Original) The isolated nucleic acid molecule of claim 15 wherein the heterologous protein destabilization sequence is a PEST sequence.

33. (Original) The isolated nucleic acid molecule of claim 15 wherein the heterologous protein destabilization sequence is from the C-terminus of a mammalian ornithine decarboxylase.

34. (Original) The isolated nucleic acid molecule of claim 15 wherein the heterologous protein destabilization sequence is CL1, CL2, CL6, CL9, CL10, CL11, CL12, CL15, CL16, CL17 or SL17.

35. (Original) A vector comprising the nucleic acid molecule of claim 1, 2 or 3.

36. (Original) The vector of claim 35 wherein the nucleic acid molecule is operably linked to a regulatable promoter.

37. (Original) The vector of claim 36 wherein the promoter is a repressible promoter.

38. (Original) The vector of claim 34 wherein the nucleic acid molecule comprises SEQ ID NO:49, SEQ ID NO:75, SEQ ID NO:76, SEQ ID NO:77, SEQ ID NO:78, SEQ ID NO:79, SEQ ID NO:80 or a fragment thereof that encodes a fusion polypeptide with substantially the same activity as the corresponding full-length fusion polypeptide encoded by SEQ ID NO:49, SEQ ID NO:75, SEQ ID NO:76, SEQ ID NO:77, SEQ ID NO:78, SEQ ID NO:79 or SEQ ID NO:80.

39. (Original) A fusion polypeptide encoded by the nucleic acid molecule of claim 1, 2 or 3.

40. (Original) The fusion polypeptide of claim 38 wherein the reporter protein is chloramphenicol acetyltransferase, luciferase, beta-glucuronidase or beta-galactosidase.

41. (Original) A host cell comprising the vector of claim 35.

42. (Original) The host cell of claim 41 which is stably transfected with the vector that encodes a fusion polypeptide comprising a luminescent protein.

43. (Original) The host cell of claim 42 wherein the signal emitted by the host cell comprising the vector is greater than the signal emitted by a corresponding host cell comprising a vector which lacks one or more of the destabilization sequences.

44. (Original) A stable cell line comprising the vector of claim 35 wherein the signal emitted by the reporter protein is equal to or greater than a signal emitted by a corresponding stable cell line comprising a vector which lacks one or more of the heterologous destabilization sequences.

45. (Original) A method to detect a reporter protein in a cell, comprising:

a) contacting a cell with the vector of claim 35; and

b) detecting or determining the presence or amount of the reporter protein in the cell or a lysate thereof.